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EXAMINER
FORMAN, BETTY J

ART UNIT	PAPER NUMBER
1634	13

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Please find below and/or attached an Office communication concerning this application or proceeding.

Re-mailed

Office Action Summary	Application No.	Applicant(s)	
	09/528,014	BARANY ET AL.	
	Examiner	Art Unit	
	BJ Forman	1634	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE *3* MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *15 November 2001* .

2a)☒ This action is **FINAL**. 2b)☐ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-22* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-22* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

11)☐ The proposed drawing correction filed on _____ is: a)☐ approved b)☐ disapproved by the Examiner.

If approved, corrected drawings are required in reply to this Office action.

12)☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. §§ 119 and 120

13)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____ .

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

14)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application).

a) ☐ The translation of the foreign language provisional application has been received.

15)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO-1449) Paper No(s) *12* .
4) ☐ Interview Summary (PTO-413) Paper No(s). _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: .

DETAILED ACTION

1. This action is in response to papers filed 15 November 2001 in Paper No. 11 in which claims 1-14 and 17 were amended. All of the amendments have been thoroughly reviewed and entered. The previous rejections in the Office Action of Paper No. 8 dated 15 May 2001 under 35 U.S.C. 112, second paragraph are withdrawn in view of the amendments. The previous rejections under 35 U.S.C. 103(a) are maintained. All of the arguments have been thoroughly reviewed and are discussed below.

New grounds for rejection necessitated by the Information Disclosure Statement filed 3 December 2001 are discussed.

The examiner's Art Unit has changed from 1655 to 1634. Please address future correspondence to Art Unit 1634.

Currently claims 1-17 are under prosecution.

Claim Rejections - 35 USC § 103

2. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

3. Claims 1, 3-7 & 17 are rejected under 35 U.S.C. 103(a) as being unpatentable over Barany et al. (WO 97/31256, published 28 August 1997) in view of Jacobson et al. (Oncogene, 1994, 9: 553-563).

Regarding Claim 1, Barany et al. teach a method for identifying one or more sequences differing by one or more single-base changes, insertions or deletion in a plurality of target nucleotide sequences in a sample (page 10, lines 23-26) comprising: blending the sample, primary oligonucleotide primers and polymerase and performing a polymerase chain reaction (PCR) to produce primary extension products; providing a plurality of probe sets wherein a first probe, having an extension-specific portion and a detectable label and a second probe having an extension-specific portion wherein the probe of a set ligate together when hybridized adjacent to one another on a complementary extension product-specific portion; and detecting the labels of the ligation products to thereby identify the presence of one or more target sequences in the sample (page 15, line 16-page 16, line 29). Barany et al. do not teach the method wherein after the PCR, a second PCR is performed to produce secondary PCR products comprising a restriction enzyme site followed by restriction enzyme digestion and a third PCR. Jacobson et al. teach a similar method comprising: blending the sample, primary oligonucleotide primers and polymerase and performing PCR to produce primary extension products; blending the primary extension products, secondary primers to produce secondary extension products; subjecting the secondary extension products to an endonuclease digestion reaction thus destroying the high abundance secondary extension products; and performing a third PCR (page 554, Fig. 1) wherein the secondary primers have target-specific portions and produce secondary PCR products comprising a restriction enzyme site (page 554, Fig. 1, step 2) wherein the method identifies one or more low abundance sequences differing by one or more single-base changes (K-ras mutant) from a high abundance sequence (wild type) in a plurality of target sequences (page 555, left column, second full paragraph). It would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to modify the single PCR amplification of Barany et al. by adding a secondary PCR to incorporate a restriction enzyme site into the PCR products as taught by Jacobson et al. for the expected benefit of producing a higher quantity of sequence-specific sequences and to thereby detect

and identify a low abundance sequence of interest within a sample containing high abundance sequences as taught by Jacobson et al. (page 555, left column, second full paragraph).

Regarding Claim 3, Barany et al. teach the method wherein a probe of each probe set has an addressable array-specific portion said method further comprising: providing a solid support with different capture oligonucleotides immobilized at different sites wherein the capture oligonucleotides have sequences complementary to the addressable array-specific portions; and contacting the ligation products with the solid support to capture the addressable array-specific portions to the solid support; and detecting the presence of ligation product immobilized to the solid support (page 11, lines 1-16).

Regarding Claim 4, Barany et al. teach the method wherein the relative amounts of the one or more sequences is quantified by comparing the amount of ligation product sequences generated to ligation products generated from known amounts of marker sequences (page 19, lines (page 19, lines 16-34).

Regarding Claim 5, Barany et al. teach as few as a single molecule can be detected (page 22, lines 33-37) but they do not teach the molar ratio of low to high abundance sequences. However, Jacobson et al. teach the similar method wherein the relative amounts of low abundance to high abundance ratio is less than 1:1,000 (page 555, left column second full paragraph, lines 8-10).

Regarding Claim 6, Barany et al. teach as few as a single molecule can be detected (page 22, lines 33-37) but they do not teach the molar ratio of low to high abundance sequences. However, Jacobson et al. teach the similar method wherein the relative amounts of low abundance to high abundance ratio is less than 1:10,000 (page 555, left column second full paragraph, lines 8-10).

Regarding Claim 7, Barany et al. teach as few as a single molecule can be detected (page 22, lines 33-37) but they do not teach the molar ratio of low to high abundance sequences. However, Jacobson et al. teach the similar method wherein the relative amounts of

low abundance to high abundance ratio is less than 1:100,000 (page 555, left column second full paragraph, lines 8-10).

Regarding Claim 17, Barany et al. do not teach including a restriction enzyme digestion step. However, Jacobson et al. teach the similar method wherein the restriction enzyme digestion is repeated to selectively destroy the high abundance extension products. It would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to have included the restriction enzyme digestion taught by Jacobson et al. and to repeat the digestion to thereby enrich for the sequence of interest and detect a low abundance sequence within a sample comprising high abundance sequences as taught by Jacobson et al. (page 555, left column, second full paragraph).

Response to Arguments

4. Applicant's comments on pages 9-16 of the Response regarding "Cancer Detection", "Detection of Minority Nucleic Acid Sequences", "Nucleotide Conversion Fidelity" and "Optimization of PCR/RE/LDR" are noted. However, because the comments do not address the pending claims and rejections, the comments are not addressed by the examiner.

Applicant argues that Barany does not suggest using the claimed steps of subjecting the primary polymerase chain reaction mixture to two or more polymerase chain reaction cycles, subjecting the secondary polymerase chain reaction mixture to two or more polymerase chain reaction cycles, or subjecting the endonuclease digestion reaction mixture to an endonuclease digestion reaction. For the record, it is noted that Barany does teach subjecting the primary polymerase chain reaction mixture to two or more polymerase chain reaction cycles (page 15, lines 28-29). Applicant further argues that Jacobson does not teach using the ligase detection reaction in conjunction with their method and they do not teach their method is useful or capable of being incorporated into another detection methodology. In response to applicant's arguments against the references individually, one cannot show nonobviousness by attacking references individually where the rejections are based on combinations of references. See *In re Keller*, 642 F.2d 413, 208 USPQ 871 (CCPA 1981); *In re Merck & Co.*, 800 F.2d 1091, 231 USPQ 375 (Fed. Cir. 1986). In response to applicant's argument that there is no suggestion to combine the references, the examiner recognizes that obviousness can only be established by combining or modifying the teachings of the prior art to produce the claimed invention where there is some teaching, suggestion, or motivation to do so found either in the references themselves or in the knowledge generally available to one of ordinary skill in the art.

See *In re Fine*, 837 F.2d 1071, 5 USPQ2d 1596 (Fed. Cir. 1988)and *In re Jones*, 958 F.2d 347, 21 USPQ2d 1941 (Fed. Cir. 1992). In this case, Jacobson specifically teaches that their PCR method incorporating a restriction enzyme digest step followed by further PCR is "extremely sensitive" compared to other PCR-derivatized methods (page 557, left column, first paragraph). This teaching clearly suggests that incorporation of a restriction enzyme digestion step into PCR-derived reactions would increase the sensitivity of those PCR reactions. Therefore, one of skill in the art would have been motivated by this suggestion of Jacobson to incorporate a restriction enzyme site into PCR reactions and/or PCR-derived reactions for the expected benefit of increased sensitivity as taught by Jacobson et al. (page 555, left column, second full paragraph). Specifically, by incorporation of the restriction enzyme step, one skilled in the art would expected to produce a higher quantity of sequence-specific sequences to thereby detect and identify a low abundance sequence of interest within a sample containing high abundance sequences as taught by Jacobson et al. (page 555, left column, second full paragraph).

Applicant argues that Barany and Jacobson are not properly combinable because Barany teaches that their method effectively identifies a single base difference and does not indicate that further procedures are desirable or needed. The argument has been considered but is not found persuasive because as stated in the paragraph above, Jacobson teaches the advantages of adding the restriction enzyme digestion step to other PCR-derived methods i.e. extreme sensitivity. Therefore, the skilled practitioner in the art would have been motivated to modify the method of Barany by incorporating a restriction enzyme digestion step for the obvious benefit of increasing assay sensitivity as suggested by Jacobson (page 555, left column, second full paragraph).

Applicant argues that the office action's suggestion that utilizing the features of Jacobson in conjunction with Barany's process would be beneficial in permitting the detection and identification of low abundance sequences is entirely speculative and based on nothing more than hindsight. In response to applicant's argument that the examiner's conclusion of obviousness is based upon improper hindsight reasoning, it must be recognized that any judgment on obviousness is in a sense necessarily a reconstruction based upon hindsight reasoning. But so long as it takes into account only knowledge which was within the level of ordinary skill at the time the claimed invention was made, and does not include knowledge gleaned only from the applicant's disclosure, such a reconstruction is proper. See *In re McLaughlin*, 443 F.2d 1392, 170 USPQ 209 (CCPA 1971). In response to applicant's argument that the examiner's conclusion is based on speculation, the argument is not found persuasive because the examiner has pointed to the teaching of Jacobson which clearly teaches that incorporation of a restriction enzyme digestion step improves sensitivity of PCR-derived

methods (page 555, left column, second full paragraph). This teaching clearly suggests other PCR methods would be more sensitive if the restriction enzyme digestion step was incorporated.

Applicant argues that the method of Jacobson has been shown to be susceptible to a high degree of false positives which makes it unsuitable for the purpose of the present invention. Applicant provides 3 exhibits to support the argument. The argument and exhibits have been considered but are not found persuasive for numerous reasons. First, as discussed above, Jacobson clearly teaches the advantages of modifying PCR reactions by incorporating their restriction enzyme digestion step. Second, the teaching of Jacobson is prior art and one skilled in the art would have been motivated by the teaching of Jacobson to modify a PCR reaction by incorporating a restriction enzyme digestion step for the obvious benefits of extreme sensitivity (page 555, left column, second full paragraph). Third, Barany specifically teaches that their ligation step following the PCR reactions eliminates the problems of multiplex PCR by i.e. mismatch priming because their ligation reaction is base-specific (page 7, line 32-page 3). Therefore, the skilled practitioner would have been motivated to increase sensitivity by incorporation a restriction enzyme digestion step following the PCR amplification and prior to the ligation because one skilled in the art would have known that any product resulting from mismatch priming would be eliminated from detection by the subsequent ligation step in the method of Barany (page 34-40).

Finally, none of the exhibits specifically address, discuss and/or question the method of Jacobson in the 1994 reference. Therefore, the arguments regarding the susceptibility to false positives in the Jacobson method are not found persuasive because the arguments are not supported by appropriate evidence.

The arguments of counsel cannot take the place of evidence in the record. In re Schulze, 346 F.2d 600, 602, 145 USPQ 716, 718 (CCPA 1965); In re Geisler, 116 F.3d 1465, 43 USPQ2d 1362 (Fed. Cir. 1997) ("An assertion of what seems to follow from common experience is just attorney argument and not the kind of factual evidence that is required to rebut a prima facie case of obviousness."). See MPEP § 716.01(c) for examples of attorney statements which are not evidence and which must be supported by an appropriate affidavit or declaration (see MPEP 2145).

Applicant argues that the rejection of Claim 17 should be withdrawn because Barany does not teach a restriction enzyme digest and Jacobson does not teach restriction enzyme digest combined with ligation; neither Barany or Jacobson teach blending ligation products with restriction enzyme; and Jacobson does not suggest combining their restriction digest with or following a ligation step. In response to applicant's argument that there is no suggestion to combine the references, the examiner recognizes that obviousness can only be established by combining or modifying the teachings of the prior art to produce the claimed invention where

there is some teaching, suggestion, or motivation to do so found either in the references themselves or in the knowledge generally available to one of ordinary skill in the art. See *In re Fine*, 837 F.2d 1071, 5 USPQ2d 1596 (Fed. Cir. 1988)and *In re Jones*, 958 F.2d 347, 21 USPQ2d 1941 (Fed. Cir. 1992). In this case, as stated above, Jacobson specifically teach that their PCR method incorporating a restriction enzyme digest step followed by further PCR is "extremely sensitive" compared to other PCR derivatized methods (page 557, left column, first paragraph). This teaching clearly suggests that incorporation of a restriction enzyme digestion step increases sensitivity of PCR reactions. Therefore, one of skill in the art would have been motivated by this suggestion of Jacobson to incorporate a restriction enzyme site into PCR reactions and/or PCR derived reactions for the expected benefit providing an extremely sensitive assay (Jacobson et al., page 555, left column, second full paragraph).

5. Claim 2 is rejected under 35 U.S.C. 103(a) as being unpatentable over Barany et al. (WO 97/31256, published 28 August 1997) in view of Jacobson et al. (Oncogene, 1994, 9: 553-563) as applied to claims 1, 3-7 and 17 above and further in view of Day et al. (Genomics, 1995, 29: 152-162).

Regarding Claim 2, Barany et al. teach the method for identifying one or more sequences differing by one or more single-base changes, insertions or deletion in a plurality of target nucleotide sequences in a sample (page 10, lines 23-26) wherein the probes in a particular set have are designed so that the ligation product sequences which they form are distinguished from other sequences: wherein the ligation products are separated and distinguished (page 14, lines 26-38) but they do not teach the ligation products are separated and distinguished by electrophoretic mobility. However, Day et al. teach a similar method comprising: blending the sample, primary oligonucleotide primers and polymerase and performing a polymerase chain reaction (PCR) to produce primary extension products; providing a plurality of probe sets wherein a first probe, having an extension-specific portion and a detectable label and a second probe having an extension-specific portion wherein the probe of a set ligate together when hybridized adjacent to one another on a complementary

extension product-specific portion; and detecting the labels of the ligation products to thereby identify the presence of one or more target sequences in the sample wherein the primers in a particular probe set have a unique length (page 157, Fig. 2) whereby ligation products are separating and distinguishing by electrophoretic mobility (page 154, left column, third full paragraph). It would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to modify the method of Barany so as to detect the ligation product using the electrophoretic mobility assay taught by Day et al. for the expected benefits of speed, sensitivity and accuracy i.e. identifying ligation products within a single multiplex reaction by using an automated sequencer and sequencing software as taught by Day et al. (page 161, left column last paragraph).

Response to Arguments

6. Applicant traverses the above rejection because Day does not overcome the deficiencies of Barany and Jacobson as argued above. The argument has been considered but is not found persuasive for the reasons stated above, see ¶ 4.

7. Claims 8 and 10-16 are rejected under 35 U.S.C. 103(a) as being unpatentable over Barany et al. (WO 97/31256, published 28 August 1997) in view of Jacobson et al. (Oncogene, 1994, 9: 553-563) as applied to claims 1, 3-7 and 17 above and further in view of Cook et al. (U.S. Patent No. 5,859,221, filed 6 June 1995).

Regarding Claim 8, Barany et al. teach the method for identifying one or more sequences differing by one or more single-base changes, insertions or deletion in a plurality of target nucleotide sequences in a sample (page 10, lines 23-26) wherein the primers comprise nucleotide analogs (page 13, lines 35-38) but they do not teach the method comprising a pre-secondary oligonucleotide primer set wherein at least one primer contains one or more nucleotide analogs. However, multiple PCR amplifications nucleotide analogs were well known in the art at the time the claimed invention was made. Specifically, Cook et al. teach that sequences comprising nucleotide analogs resist nuclease degradation and hybridize with

greater fidelity (Column 4, lines 38-42). It would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to modify the primer of Barany et al. to comprise one or more nucleotide analogs for the expected benefits of resistance to nuclease degradation and greater fidelity of hybridization as taught by Cook et al. (Column 4, lines 38-42). The skilled practitioner in the art would have been further motivated to modify the PCR amplifications of Barany et al. and Jacobson et al. to perform a pre-secondary amplification for the known benefits of PCR i.e. sensitivity and increased product yield as taught by Barany et al. (page 7, lines 32-35).

Regarding Claim 10, Barany et al. teach the method wherein a probe of each probe set has an addressable array-specific portion said method further comprising: providing a solid support with different capture oligonucleotides immobilized at different sites wherein the capture oligonucleotides have sequences complementary to the addressable array-specific portions; and contacting the ligation products with the solid support to capture the addressable array-specific portions to the solid support; and detecting the presence of ligation product immobilized to the solid support (page 11, lines 1-16).

Regarding Claim 11, Barany et al. teach the method wherein the relative amounts of the one or more sequences is quantified by comparing the amount of ligation product sequences generated to ligation products generated from known amounts of marker sequences (page 19, lines (page 19, lines 16-34).

Regarding Claim 12, Barany et al. teach as few as a single molecule can be detected (page 22, lines 33-37) but they do not teach the molar ratio of low to high abundance sequences. However, Jacobson et al. teach the similar method wherein the relative amounts of low abundance to high abundance ratio is less than 1:1,000 (page 555, left column second full paragraph, lines 8-10).

Regarding Claim 13, Barany et al. teach as few as a single molecule can be detected (page 22, lines 33-37) but they do not teach the molar ratio of low to high abundance

sequences. However, Jacobson et al. teach the similar method wherein the relative amounts of low abundance to high abundance ratio is less than 1:10,000 (page 555, left column second full paragraph, lines 8-10).

Regarding Claim 14, Barany et al. teach as few as a single molecule can be detected (page 22, lines 33-37) but they do not teach the molar ratio of low to high abundance sequences. However, Jacobson et al. teach the similar method wherein the relative amounts of low abundance to high abundance ratio is less than 1:100,000 (page 555, left column second full paragraph, lines 8-10).

Regarding Claim 15, Barany et al. and Cook et al. do not teach the analog of at least one primer is at the 3' end of the primer. However, it would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to apply the teaching of Cook et al. wherein the analogs resist nuclease digestion and hybridize with greater fidelity to the primers of Barany et al. and Jacobson et al. and to place an analog at the 3' end of at least on primer for the expected benefit of preventing digestion from nuclease which are known in the art to digest nucleic acid sequences from the 3' end and for the expected benefit of increasing 3' end hybridization fidelity to thereby improve PCR primer extension which is known in the art to depend upon the 3' primer-template hybridization.

Regarding Claim 16, Barany et al. do not teach the claimed nucleotide analogs. However, Cook et al. teach the claimed analogs (Column 6, lines 8-29). It would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to modify the primer of Barany et al. to comprise one or more nucleotide analogs for the expected benefits of resistance to nuclease degradation and greater fidelity of hybridization as taught by Cook et al. (Column 4, lines 38-42).

Response to Arguments

8. Applicant traverses the above rejection because Cook does not teach identifying one or more low abundance sequences differing by one or more single base changes, insertions, or deletions from a high abundance sequence utilizing a ligase detection reaction as in Barany

and there is not suggestion in Cook that their modified oligonucleotides can be utilized in conjunction with an endonuclease reaction as in Jacobson and therefore, Application argues, there would have been no reason to combine the teaching of Cook with that of Jacobson and/or Barany. In response to applicant's argument that there is no suggestion to combine the references, the examiner recognizes that obviousness can only be established by combining or modifying the teachings of the prior art to produce the claimed invention where there is some teaching, suggestion, or motivation to do so found either in the references themselves or in the knowledge generally available to one of ordinary skill in the art. See *In re Fine*, 837 F.2d 1071, 5 USPQ2d 1596 (Fed. Cir. 1988)and *In re Jones*, 958 F.2d 347, 21 USPQ2d 1941 (Fed. Cir. 1992). In this case, Cook teaches that their modified oligonucleotides display greater fidelity of hybridization than any other known oligonucleotide (Column 4, lines 38-42). Cook teaches that their modified oligonucleotides are resistant to nuclease degradation (Column 4, lines 8-13). Additionally, Cook teaches that oligonucleotides that are nuclease resistant and fidelity of hybridization are of "great importance" as research reagents (Column 3, lines 4-8). Therefore, the skilled practitioner in the art would have been motivated to modify the primers of Barany with the nucleotide analogs of Cook to thereby provide primers having the highest fidelity of hybridization in addition to being nuclease resistant based on the importance of fidelity and resistance as taught by Cook (Column 3, lines 4-8). Additionally, one skilled in the art would have been motivated to modify the Barany and/or Jacobson primers with the nucleotide analogs of Cook thereby providing primers having the highest fidelity of hybridization for the obvious benefit of obtaining the highest fidelity of primer-target binding (Column 4, lines 38-42).

9. Claim 9 is rejected under 35 U.S.C. 103(a) as being unpatentable over Barany et al. (WO 97/31256, published 28 August 1997) in view of Jacobson et al. (Oncogene, 1994, 9: 553-563) and Cook et al. (U.S. Patent No. 5,859,221, filed 6 June 1995) as applied to claims 8 and 10-16 above and further in view of Day et al. (Genomics, 1995, 29: 152-162).

Regarding Claim 9, Barany et al. teach the method for identifying one or more sequences differing by one or more single-base changes, insertions or deletion in a plurality of target nucleotide sequences in a sample (page 10, lines 23-26) wherein the ligation products are separated and distinguished (page 14, lines 26-38) but they do not teach the ligation

products are separated and distinguished by electrophoretic mobility. However, Day et al. teach a similar method comprising: blending the sample, primary oligonucleotide primers and polymerase and performing a polymerase chain reaction (PCR) to produce primary extension products; providing a plurality of probe sets wherein a first probe, having an extension-specific portion and a detectable label and a second probe having an extension-specific portion wherein the probe of a set ligate together when hybridized adjacent to one another on a complementary extension product-specific portion; and detecting the labels of the ligation products to thereby identify the presence of one or more target sequences in the sample wherein the primers in a particular probe set have a unique length (page 157, Fig. 2) whereby ligation products are separating and distinguishing by electrophoretic mobility (page 154, left column, third full paragraph). It would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to modify the ligation product identification of Barany et al. with the electrophoretic mobility identification taught by Day et al. for the expected benefits of with speed, sensitivity and accuracy i.e. identifying ligation products within a single multiplex reaction by using an automated sequencer and sequencing software as taught by Day et al. (page 161, left column last paragraph).

Response to Arguments

10. Applicant traverses the above rejection because Day does not overcome the deficiencies of Barany and Jacobson as argued above. The argument has been considered but is not found persuasive for the reasons stated above, see ¶ 4.

11. Claims 1, 2, 5-7 & 17 are rejected under 35 U.S.C. 103(a) as being unpatentable over Belgrader et al. (Genome Science & Technology, 1996, 1(2): 77-87) in view of Jacobson et al. (Oncogene, 1994, 9: 553-563).

Regarding Claim 1, Belgrader et al. teach a method for identifying one or more sequences differing by one or more single-base changes, insertions or deletion in a plurality of target nucleotide sequences in a sample (comprising: blending the sample, primary oligonucleotide primers and polymerase and performing a polymerase chain reaction (PCR) to produce primary extension products; blending the primary extension products, secondary oligonucleotide primers and polymerase and performing a second polymerase chain reaction (PCR) to produce secondary extension products; providing a plurality of probe sets wherein a first probe, having an extension-specific portion and a detectable label and a second probe having an extension-specific portion wherein the probe of a set ligate together when hybridized adjacent to one another on a complementary extension product-specific portion; and detecting the labels of the ligation products to thereby identify the presence of one or more target sequences in the sample (page 79 and page 80, Fig. 1). Belgrader et al. do not teach the method comprising producing secondary PCR products comprising a restriction enzyme site followed by restriction enzyme digestion and a third PCR. Jacobson et al. teach a similar method comprising: blending the sample, primary oligonucleotide primers and polymerase and performing PCR to produce primary extension products; blending the primary extension products, secondary primers to produce secondary extension products; subjecting the secondary extension products to an endonuclease digestion reaction thus destroying the high abundance secondary extension products; and performing a third PCR (page 554, Fig. 1) wherein the secondary primers have target-specific portions and produce secondary PCR products comprising a restriction enzyme site (page 554, Fig. 1, step 2) wherein the method identifies one or more low abundance sequences differing by one or more single-base changes (K-ras mutant) from a high abundance sequence (wild type) in a plurality of target sequences (page 555, left column, second full paragraph). It would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to modify the single PCR amplification of Belgrader et al. by adding a secondary PCR to incorporate a restriction enzyme

site into the PCR products as taught by Jacobson et al. for the expected benefit of producing a higher quantity of sequence-specific sequences and to thereby detect and identify a low abundance sequence of interest within a sample containing high abundance sequences as taught by Jacobson et al. (page 555, left column, second full paragraph).

Regarding Claim 2, Belgrader et al teach the method wherein the oligonucleotide probes have a unique length whereby the ligation product sequences which they form are distinguished from other ligation products by electrophoretic mobility prior to detection (page 83, first and second paragraphs and Fig. 2).

Regarding Claim 5, Belgrader et al. teach the method wherein one or more low abundance sequences is present in less than a 1:1,000 ration relative to the amount of high abundance sequence (Abstract).

Regarding Claim 6, Belgrader et al. teach the method wherein one or more low abundance sequences is present in less than a 1:10,000 ration relative to the amount of high abundance sequence (Abstract).

Regarding Claim 7, Belgrader et al. teach the method wherein one or more low abundance sequences is present in less than a 1:100,000 ration relative to the amount of high abundance sequence (Abstract).

Regarding Claim 17, Belgrader et al. do not teach including a restriction enzyme digestion step. However, Jacobson et al. teach the similar method wherein the restriction enzyme digestion is repeated to selectively destroy the high abundance extension products. It would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to have included the restriction enzyme digestion taught by Jacobson et al. and to repeat the digestion to thereby enrich for the sequence of interest and detect a low abundance sequence within a sample comprising high abundance sequences as taught by Jacobson et al. (page 555, left column, second full paragraph).

12. Claims 8, 9 and 12-16 are rejected under 35 U.S.C. 103(a) as being unpatentable over Belgrader et al. (Genome Science & Technology, 1996, 1(2): 77-87) in view of Jacobson et al. (Oncogene, 1994, 9: 553-563) as applied to claims 1, 3-7 and 17 above and further in view of Cook et al. (U.S. Patent No. 5,859,221, filed 6 June 1995).

Regarding Claim 8, Belgrader et al. teach the method for identifying one or more sequences differing by one or more single-base changes, insertions or deletion in a plurality of target nucleotide sequences in a sample (Abstract) but they do not teach the method comprising a pre-secondary oligonucleotide primer set wherein at least one primer contains one or more nucleotide analogs. However, multiple PCR amplifications nucleotide analogs were well known in the art at the time the claimed invention was made. Specifically, Cook et al. teach that sequences comprising nucleotide analogs resist nuclease degradation and hybridize with greater fidelity (Column 4, lines 38-42). It would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to modify the primer of Belgrader et al. to comprise one or more nucleotide analogs for the expected benefits of resistance to nuclease degradation and greater fidelity of hybridization as taught by Cook et al. (Column 4, lines 38-42).

Regarding Claim 9, Belgrader et al. teach the method for identifying one or more sequences differing by one or more single-base changes, insertions or deletion in a plurality of target nucleotide sequences in a sample wherein the ligation products are separated and distinguished wherein the ligation products are separated and distinguished by electrophoretic mobility (page 82, first paragraph and Fig. 2).

Regarding Claim 12, Belgrader et al. teach the method wherein one or more low abundance sequences is present in less than a 1:1,000 ration relative to the amount of high abundance sequence (Abstract).

Regarding Claim 13, Belgrader et al. teach the method wherein one or more low abundance sequences is present in less than a 1:10,000 ration relative to the amount of high abundance sequence (Abstract).

Regarding Claim 14, Belgrader et al. teach the method wherein one or more low abundance sequences is present in less than a 1:100,000 ration relative to the amount of high abundance sequence (Abstract).

Regarding Claim 15, Belgrader et al. and Cook et al. do not teach an analog of at least one primer is at the 3' end of the primer. However, it would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to apply the teaching of Cook et al. wherein the analogs resist nuclease digestion and hybridize with greater fidelity to the primers of Belgrader et al. and Jacobson et al. and to place an analog at the 3' end of at least on primer for the expected benefit of preventing digestion from nuclease which are known in the art to digest nucleic acid sequences from the 3' end and for the expected benefit of increasing 3' end hybridization fidelity to thereby improve PCR primer extension which is known in the art to depend upon the 3' primer-template hybridization.

Regarding Claim 16, Belgrader et al. do not teach the claimed nucleotide analogs. However, Cook et al. teach the claimed analogs (Column 6, lines 8-29). It would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to modify the primer of Belgrader et al. to comprise one or more nucleotide analogs for the expected benefits of resistance to nuclease degradation and greater fidelity of hybridization as taught by Cook et al. (Column 4, lines 38-42).

13. Applicant's amendment necessitated the new ground(s) of rejection presented in this Office action. Accordingly, **THIS ACTION IS MADE FINAL**. See MPEP § 706.07(a). Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the date of this final action.

Conclusion

14. No claim is allowed.

15. Any inquiry concerning this communication or earlier communications from the examiner should be directed to BJ Forman whose telephone number is (703) 306-5878. The examiner can normally be reached on 6:30 TO 4:00.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Gary Jones can be reached on (703) 308-1152. The fax phone numbers for the organization where this application or proceeding is assigned are (703) 308-4242 for regular communications and (703) 308-8724 for After Final communications.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is (703) 308-0196.

BJ Forman, Ph.D.
Patent Examiner
Art Unit: 1634
March 14, 2002

W. Gary Jones
Supervisory Patent Examiner
Technology Center 1600